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# Four new East Asian species of *Aleurodiscus* with echinulate basidiospores

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## Abstract

Four new species of *Aleurodiscus* sensu lato with echinulate basidiospores are described from East Asia: *A. alpinus*, *A. pinicola*, *A. senticosus*, and *A. sichuanensis*. *Aleurodiscus alpinus* is from northwest Yunnan of China where it occurs on *Rhododendron* in montane habitats. *Aleurodiscus pinicola* occurs on *Pinus* in montane settings in Taiwan and northwest Yunnan. *Aleurodiscus senticosus* is from subtropical Taiwan, where it occurs on angiosperms. *Aleurodiscus sichuanensis* is reported from southwest China on angiosperms in montane environments. Phylogenetic relationships of these four new species were inferred from analyses of a combined dataset consisting of three genetic markers, viz. 28S, nuc rDNA ITS1-5.8S-ITS2 (ITS), and a portion of the translation elongation factor 1-alpha gene, *TEF1*.

**Keywords**

China, corticioid fungi, Taiwan, taxonomy, wood-decaying fungi

## Introduction

The genus *Aleurodiscus* Rabenh. ex J. Schröt. belongs to the Stereaceae Pilát of the Russulales Kreisel ex P.M. Kirk, P.F. Cannon & J.C. David. However, whether to keep *Aleurodiscus* in a broad or a narrow sense has long been a puzzling issue in the taxonomy of Basidiomycota (Boidin et al 1985; Núñez and Ryvarden 1997; Wu et al. 2001; Larsson and Larsson 2003; Miller et al. 2006; Larsson 2007; Wu et al. 2010), because diagnostic characters are highly variable among species. *Aleurodiscus* s.l. is characterized by cupulate,

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effused or effused-reflexed basidiocarps, a monomitic or dimitic hyphal system with simple-septate or clamped generative hyphae, smooth or ornamented amyloid basidiospores, and sterile organs such as acanthophyses, gloeocystidia, hyphidia, and dendrohyphidia (Núñez and Ryvarden 1997). The characteristics used for separating segregate genera within *Aleurodiscus* s.l. (*Acanthobasidium* Oberw., *Acanthofungus* Sheng H. Wu et al, *Acanthophysellum* Parmasto, *Aleurobotrys* Boidin, *Aleurodiscus* s.s., *Aleurocystidiellum* P.A. Lemke, *Gloeosoma* Bres., and *Neoaleurodiscus* Sheng H. Wu) as well as *Stereum* Hill ex Pers. and *Xylobolus* P. Karst. were provided by Wu et al. (2001, table 1) and Wu et al. (2010, table 1). Currently, 169 names are recorded under *Aleurodiscus*, of which about 85 taxa are generally accepted worldwide (http://www.indexfungorum.org/). Since the year 2000, new species of *Aleurodiscus* s.l. have been proposed by Simpson and Grgurinovic (2003), Hjortstam et al. (2009), Ryvarden et al. (2012), Gorjón et al. (2013), Maninder et al. (2014), Dai and He (2016), Dai et al. (2017a, b), Ghobad-Nejhad and Langer (2018), and Tian et al. (2018). Since the phylogenetic relationships of the taxa in *Aleurodiscus* s.l., as well as in the Stereaceae at large, are not resolved, we adopt a broad and inclusive generic concept of *Aleurodiscus* for the new taxa presented in this study.

During a two-decade long, ongoing survey of corticioid fungi from mainland China and Taiwan, we have found four new species of *Aleurodiscus* with echinulate basidiospores based on morphological characters. In addition, phylogenetic analyses of a nuclear rDNA 28S D1–D2 domains (28S) dataset and analyses of a second dataset consisting of three genetic markers – nuc rDNA 28S D1–D2 domains (28S), nuc rDNA ITS1-5.8S-ITS2 (ITS), and translation elongation factor 1-alpha (*TEF1*) – are performed to complement our morphological observations and place the newly described species in a molecular phylogenetic framework.

## Materials and methods

### Morphological and cultural studies

Macroscopic and microscopic studies were based on dried specimens. Color names from Rayner (1970) are capitalized. Thin free-hand sections of basidiocarps were prepared for microscopic study. For observations and measurements of microscopic characters, sections were mounted in 5% KOH to ensure rehydration. A blue-black color change with Melzer's reagent (IKI) indicates an amyloid reaction. Cotton blue (CB) was used as mounting medium to determine cyanophily. Sulphoaldehyde (SA) was used to detect a sulphuric reaction of gloeocystidia; a bluish black color change with SA indicates a positive reaction. The following abbreviations are used for basidiospore measurements: L = mean spore length with standard deviation, W = mean spore width with standard deviation, Q = variation in L/W ratio, and n = number of spores measured from each specimen. Apiculi and ornamentation were excluded in spore measurements. Living mycelia were isolated from the woody substratum beneath the basidiocarps, and were cultured on 1.5% malt extract agar (MEA). Fungal specimens and living cultures used in this study are deposited in the herbaria of the National Mu-

seum of Natural Science of ROC (TNM; Taichung City, Taiwan) and Beijing Forestry University (BJFC; Beijing, China).

## DNA extraction, polymerase chain reaction (PCR), and sequencing

Dried specimens or the mycelial colonies cultured on MEA were used for DNA extraction, carried out with a Plant Genomic DNA Extraction Miniprep System (Viogene-Biotek Corp., New Taipei City, Taiwan). Liquid N and Tissue Lyser II (Qiagen, Hilden, Germany) were used to disrupt and homogenize the fungal tissues before DNA extraction process. The primer pairs ITS1/ITS4 or ITS1F/LR22 were used for the ITS region (White et al. 1990, Gardes and Bruns 1993), and LR0R/LR3 and LR0R/LR5 were used for the 28S region (Vilgalys and Hester 1990). Efdf/1953R and 983F/2218R were used to amplify a portion of the *TEF1* gene (Rehner & Buckley 2005; Matheny et al. 2007). PCR products were purified and directly sequenced by MB Mission Biotech Company (Taipei City, Taiwan). We examined the technical quality of the newly obtained sequences by comparison to entries in GenBank. Sequences were assembled using BioEdit v7.2.5 (Hall 1999). Newly obtained sequences (Supplementary Table 1) were submitted to either GenBank through the National Center for Biotechnology Information (NCBI) or DNA Data Bank of Japan (DDBJ) (Mashima et al. 2016, Benson et al. 2018).

## Alignment and phylogenetic analyses

The newly generated sequences were added to the DNA sequence dataset employed by Dai and He (2016), so far the most inclusive alignments for analyzing *Aleurodiscus* s.l. based on three genetic markers. To achieve a comprehensive analysis, we also added some related taxa of the genera *Boidinia* Stalpers & Hjortstam, *Conferticium* Hallenb., *Gloeocystidiellum* Donk and *Megalocystidium* Jülich to the ingroup. We tried to include the type species of the genera as far as possible (Table 1). The phylogenetic tree of the 28S+ITS+ *TEF1* dataset was inferred through Maximum likelihood (ML) and Bayesian inference (BI) methods using RAxML v. 8.2.4 (Stamatakis 2014) and MrBayes v. 3.2.6 (Ronquist et al. 2012), respectively (Ronquist et al. 2012, Stamatakis 2014). The alignments were inferred in MAFFT v. 7 using the FFT-N-i strategy for 28S and *TEF1*, and Q-INS-i strategy for ITS. For the BI analysis, the best-fit model for each alignment partition was estimated by jModelTest 2 (Darriba et al. 2012) using the Akaike information criterion (AIC). For ML bootstrapping, the extended majority-rule consensus tree criterion was specified under a GTRGAMMA model with 1000 replicates. In the BI analysis, four MCMC chains were run simultaneously from a random starting tree for ten million generations. Trees were sampled every 1000 generations resulting in 10000 trees in the posterior distribution; the first 25% trees were discarded as the burn-in. Posterior probabilities (PP) were calculated based on the post-burn-in trees. ML bootstrap values (BS) and BI posterior probability (PP) values ≥ 50% and ≥ 0.7 are indicated at the nodes of the ML tree. The final sequence alignments and the phylogenetic trees are available at TreeBASE (S23581; www.treebase.org).

**Table 1.** List of species, specimens and sequences used in this study. Sequences generated in this study are shown in boldface.

| Fungal species | Specimen or strain no. | DDBJ/GenBank/EMBL accession no. | | |
|---|---|---|---|---|
| | | ITS | 28S | *TEF1* |
| *Acanthobasidium bambusicola* | He2357 | KU559343 | KU574833 | – |
| *Acanthofungus rimosus#* | Wu9601-1 | MF043521 | AY039333 | – |
| *Acanthophysellum cerussatum* | He20120920-3 | KU559339 | KU574830 | KU992716 |
| *Aleurobotrys botryosus#* | He2712 | KX306877 | KY450788 | – |
| *Aleurocystidiellum disciforme* | He3159 | KU559340 | KU574831 | KU992721 |
| *Aleurocystidiellum subcruentatum#* | He2886 | KU559341 | KU574847 | KU992720 |
| ***Aleurodiscus alpinus*** | **Wu1407-59** | **MF043522** | **MF043527** | **–** |
| ***Aleurodiscus alpinus*** | **Wu1407-55*** | **–** | **MF043526** | **LC269190** |
| ***Aleurodiscus alpinus*** | **Wu1407-61** | **MF043523** | **MF043528** | **–** |
| *Aleurodiscus amorphus#* | Ghobad-Nejhad-2464 | KU559342 | KU574832 | KU992717 |
| *Aleurodiscus amorphus#* | KHL4240 | AF506397 | AF506397 | – |
| *Aleurodiscus bambusinus* | He 4261 | KY706207 | KY706219 | **LC430911** |
| *Aleurodiscus canadensis* | Wu 1207-90 | KY706203 | KY706225 | – |
| *Aleurodiscus dextrinoideocerussatus* | EL25-97 | AF506401 | AF506401 | – |
| *Aleurodiscus dextrinoideophyses* | He 4105 | MH109050 | KY450784 | – |
| *Aleurodiscus effusus* | He2261 | KU559344 | KU574834 | KU992719 |
| *Aleurodiscus gigasporus* | Wu 0108-15 | KY706205 | KY706213 | – |
| *Aleurodiscus grantii* | HHB-14417 | KU559363 | KU574821 | KU992708 |
| *Aleurodiscus grantii* | HHB-14418 | KU559364 | KU574822 | – |
| *Aleurodiscus isabellinus* | He 5283 | MH109052 | MH109046 | **LC430912** |
| *Aleurodiscus mesaverdense* | FP-120155 | KU559359 | KU574817 | – |
| *Aleurodiscus mirabilis* | Dai13281 | KU559350 | KU574839 | KU992711 |
| *Aleurodiscus oakesii* | He2243 | KU559352 | KU574840 | – |
| *Aleurodiscus oakesii* | HHB11890-A-sp | KU559365 | KU574823 | – |
| ***Aleurodiscus pinicola*** | **Wu1106-16** | **MF043524** | **MF043529** | **–** |
| ***Aleurodiscus pinicola*** | **Wu1308-54*** | **MF043525** | **MF043530** | **LC269191** |
| ***Aleurodiscus senticosus*** | **Wu1209-7*** | **MH596849** | **MF043531** | **LC271169** |
| ***Aleurodiscus senticosus*** | **Wu1209-9** | **MH596850** | **MF043533** | **LC269192** |
| ***Aleurodiscus senticosus*** | **Wu9610-1** | **MH596851** | **MF043532** | **LC269193** |
| ***Aleurodiscus sichuanensis*** | **Wu0010-18*** | **MH596852** | **MF043534** | **LC269194** |
| ***Aleurodiscus sichuanensis*** | He 4935 | **LC430904** | **LC430907** | – |
| *Aleurodiscus subroseus* | He 4807 | MH109054 | MH109048 | – |
| ***Aleurodiscus subroseus*** | He 4895 | **LC430903** | **LC430910** | **LC430913** |
| *Aleurodiscus tenuissimus* | He3575 | KX306880 | KX842529 | – |
| *Aleurodiscus thailandicus* | He 4099 | KY450781 | KY450782 | – |
| *Aleurodiscus tropicus* | He3830 | KX553875 | KX578720 | LC269195 |
| *Aleurodiscus verrucosporus* | He 4491 | KY450786 | KY450790 | – |
| *Aleurodiscus wakefieldiae* | He2580 | KU559353 | KU574841 | KU992710 |
| *Boidinia macrospora* | Wu9202-21 | AF506377 | AF506377 | – |
| *Conferticium heimii* | CBS321.66 | AF506381 | AF506381 | – |
| *Conferticium ravum* | NH13291 | AF506382 | AF506382 | – |
| *Gloeocystidiellum aspellum* | LIN625 | AF506432 | AF506432 | – |
| ***Gloeocystidiellum porosum#*** | **Wu 1608-176** | **LC430905** | **LC430908** | – |
| *Gloeocystidiopsis cryptacanthus* | KHL10334 | AF506442 | AF506442 | – |
| *Gloeocystidiopsis flammea#* | CBS324.66 | AF506437 | AF506437 | – |
| *Heterobasidion parviporum* | 91605 | KJ651503 | KJ651561 | KU985089 |
| *Megalocystidium chelidonium* | LodgeSJ110.1 | AF506441 | AF506441 | – |
| *Megalocystidium leucoxanthum#* | HK9808 | AF506420 | AF506420 | – |
| *Megalocystidium wakullum* | Oslo-930107 | AF506443 | AF506443 | – |
| *Neoaleurodiscus fujii#* | He2921 | KU559357 | KU574845 | KU992709 |
| *Stereum complicatum* | He2234 | KU559368 | KU574828 | KU992706 |
| *Stereum hirsutum#* | Wu 1109-127 | **LC430906** | **LC430909** | – |
| *Stereum ostrea* | SHe2067 | KU559366 | KU574826 | KU992703 |
| *Stereum sanguinolentum* | He2111 | KU559367 | KU574827 | KU992705 |
| *Xylobolus frustulatus* | He2231 | KU881905 | KU574825 | KU992704 |

* Holotype, # Generic type

## Phylogeny results

The three-marker dataset was composed of 55 taxa and 2502 sites including gaps (of which 29% were parsimony-informative): 953 characters for 28S, 949 characters for ITS and 600 characters for *TEF1*. Missing sequences were treated as missing data (Table 1). After the ML search, 1000 rapid bootstrap inferences were executed. For the BI analysis, the GTR+I+G model was chosen as the best model for the 28S and *TEF1* alignments, and GTR+G was chosen for the ITS alignment. After 2.79 million

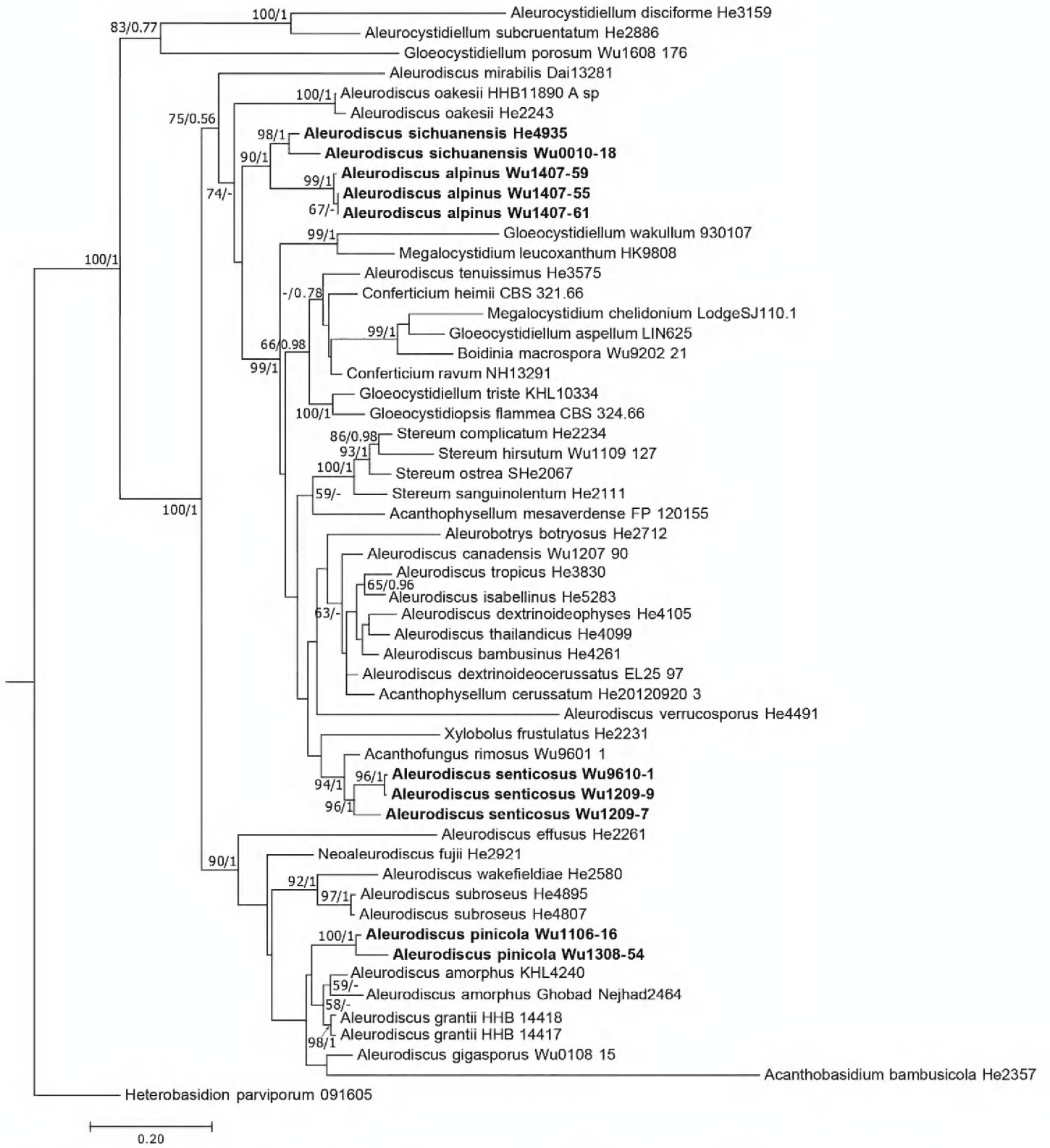


**Figure 1.** ML tree of *Aleurodiscus* and related genera of Stereaceae inferred from the 28S-ITS-*TEF1* markers. ML bootstrap values ≥ 50% and PP ≥ 0.7 from the Bayesian analysis are indicated at internodes. The presented new species are shown in boldface type.

generations, average standard deviation of split frequencies fell to 0.0099. Only the ML tree is shown given that the ML and BI analyses yielded similar topologies. The ML tree of the combined 28S, ITS and *TEF1-α* dataset (Fig. 1) showed that strains of *Aleurodiscus alpinus*, *A. pinicola*, *A. sichuanensis*, and *A. senticosus* formed separate clades in distinct lineages with high statistical support (BS = 96–100%, PP = 1). The strain of *A. sichuanensis* was sister to *A. alpinus* with significant support, BS: 90% and PP: 1.

## Taxonomy

***Aleurodiscus alpinus* Sheng H. Wu, sp. nov.**
MycoBank MB823178
Figs 2A, 3

**Typification.** CHINA. YUNNAN PROVINCE: Shangrila County, Pudacuo National Park, Bita Lake, 27°43'N, 99°58'E, 3640 m, on branch of *Rhododendron* sp., 10 Jul 2014, *S.H. Wu, Wu 1407-55* (holotype TNM F27976). GenBank 28S = MF043526, *TEF1* = LC269190.

**Etymology.** *alpinus* (L.), referring to the occurrence at high elevations.

**Diagnosis.** Resembles *Aleurodiscus cupulatus* Núñez & Ryvarden in having discoid basidiomes, clamped hyphae, similar gloeocystidia, absence of acanthophyses, branched or unbranched hyphidia, and echinulate basidiospores. *Aleurodiscus cupulatus* features much wider basidiospores than *A. alpinus*. It differs from its closest phylogenetic relative, *A. sichuanensis*, by having clamped hyphae, but lacks acanthophyses.

**Description.** Basidiomes cupuloid or discoid, solitary, occasionally fused, adnate, 350–750 μm thick in section. Hymenial surface Buff, Pale Luteous or Luteous, subceraceous, covered with crystal masses, not cracked; margin concolorous or paler, incurved, filamentous.

Hyphal system monomitic; hyphae nodose-septate. Pileus hyphae subcolorless to brownish, straight, thick-walled, walls usually thinner towards apices, usually with excreted material near apices. Subiculum uniform, with dense to compact texture, 150–500 μm thick; hyphae near substrate more or less vertical, moderately ramified, colorless, 3.5–8 μm diam, with 0.7–1.5 μm thick walls, occasionally guttulate; hyphae near hymenial layer more or less vertical, moderately ramified, colorless, fairly straight, 2.5–5 μm diam, thin- or slightly thick-walled, anastomoses occasional. Hymenial layer thickening, subhymenium differentiated from subiculum, 200–250 μm thick, with dense texture; hyphae fairly vertical, colorless, guttulate, 2–4 μm diam, thin-walled. Crystals sparsely scattered throughout section. Gloeocystidia numerous, immersed or slightly projecting, tubular, sometimes with adventitious septa near basal parts, colorless, (50–)70–200 × 4.5–12.5 μm, thin-walled, guttulate, SA+. Hyphidia numerous, sometimes branched, 40–130 × 2–6.5 μm. Basidia narrowly clavate, occasionally with one or two small protuberances, 85–165 × 16–20 μm, slightly thick-walled (ca. 0.5 μm thick), 4-sterigmate. Basidiospores ellipsoid to narrowly ellipsoid, adaxially concave, finely aculeate, thin-walled, homogenous or guttulate, amyloid, CB–, mostly 22–26 × 11–14 μm.

(22–)22.2–26(–27.8) × (11–)11.8–13.5(–14.8) μm, L = 24.2±1.7 μm, W = 12.6±2.2 μm, Q = 1.95 (n = 30) (holotype, *Wu 1407-55*); (22–)23–24.5(–26) × (10.2–)10.8–13(–14) μm, L = 23.8±1.0 μm, W = 11.8±1.0 μm, Q = 2.02 (n = 30) (*Wu 1407-59*).

**Ecology and distribution.** On dead branches of *Rhododendron* and other angiosperms at very high elevations, China, Jul.

**Additional specimens examined.** CHINA. YUNNAN PROVINCE: Shangrila County, Pudacuo National Park, Bita Lake, 27°43'N, 99°58'E, 3640 m, on branch of *Rhododendron* sp., 10 Jul 2014, *S.H. Wu*, *Wu 1407-59* (TNM F27979), *Wu 1407-61* (TNM F27981); Pudacuo National Park, 3600 m, on dead branch of *Rhododendron* sp., 28 Jul 2017, *S.H. He*, *He 4924* (BJFC), *He 4942* (BJFC); Jianchuan County, Laochunshan, 26°38'N, 99°47'E, 3400 m, on angiosperm branch, 26 Jul 2001, *S.H. Wu* & *S.Z. Chen*, *Wu 0107-22* (TNM F13507), *Wu 0107-25* (TNM F13510).

***Aleurodiscus pinicola*** **Sheng H. Wu, sp. nov.**
MycoBank MB823179
Figs 2B, 4

**Typification.** CHINA. YUNNAN PROVINCE: Lichiang, High Mountain Workstation of Kunning Botanical Garden, 27°00'N, 100°11'E, 3250 m, on branch of *Pinus densata*, 30 Aug 2013, *S.H. Wu*, *Wu 1308-54* (holotype TNM F27182). GenBank: ITS = MF043525, 28S = MF043530, *TEF1* = LC269191.

**Etymology.** *pinicola* (L.), dwelling on *Pinus*, in reference to the substrate.

**Diagnosis.** *Aleurodiscus pinicola* and *Acanthobasidium penicillatus* Burt share the features of moniliform gloeocystidia, acanthophyses with apical spines, dendrohyphidia, basidia with lateral protuberances, and aculeate basidiospores; the latter, however, has clamped hyphae and narrower basidiospores 18–27 × 12–14 (–17) μm. *Aleurodiscus pinicola* also resembles *A. oakesii* (Berk. & M.A. Curtis) Pat., however, the latter occurs on deciduous trees and has smaller basidiospores (15–20 × 13–17 μm).

Basidiomes discoid, each one up to 3.5 × 3 mm, adnate, membranaceous-subceraceous, 180–400 μm thick in section. Hymenial surface Buff or Pale Luteous, smooth, occasionally cracked; margin whitish, incurved, filamentous.

Hyphal system monomitic; most hyphae simple-septate, a few hyphal septa in junction of hymenium and subiculum with clamp connections. Subiculum uniform, with fairly dense dense texture, 60–160 μm thick; hyphae more or less vertical at resupinate parts, ± horizontal at marginal curved parts, moderately ramified, more or less interwoven, colorless, 2.5–6 μm diam, slightly thick-walled or containing thick walls up to 2 μm thick, sometimes with small oily drops, anastomoses occasional, some basal hyphae brownish yellow, with thicker walls than those elsewhere. Hymenial layer thickening, subhymenium more or less differentiated from subiculum, with dense texture, 100–270 μm thick; hyphae more or less vertical, colorless, sometimes with a short branch, usually containing minute oily drops, 2.5–5.5 μm diam, thin-walled. Crystal masses scattered throughout hymenial layer. Gloeocystidia numerous, mostly immersed or slightly projecting, cylindrical, usually strongly moniliform toward apices, usually forked, with

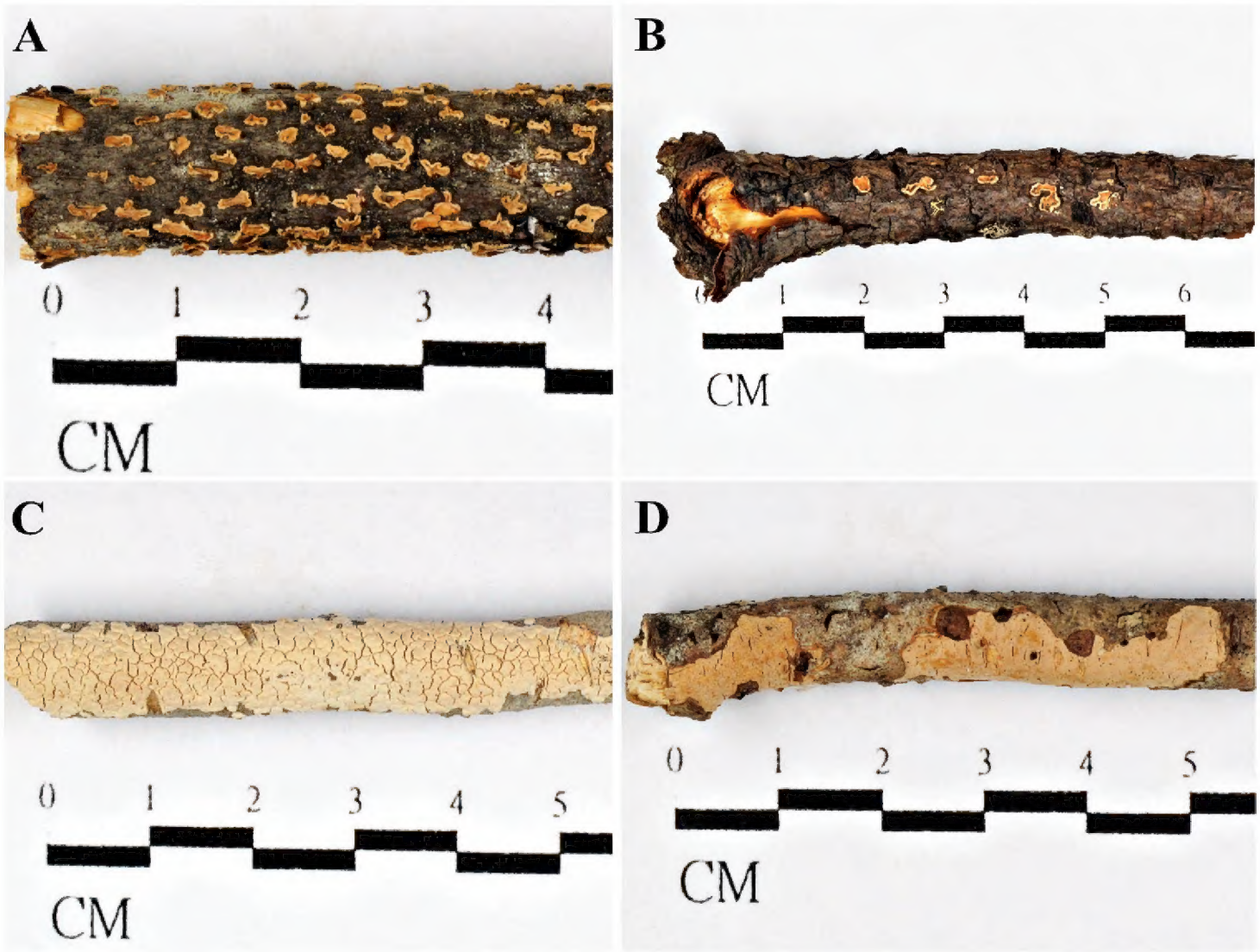


**Figure 2.** Basidiocarps **A** *Aleurodiscus alpinus* (holotype, *Wu 1407-55*) **B** *A. pinicola* (holotype, *Wu 1308-54*) **C** *A. senticosus* (holotype, *Wu 1308-54*) **D** *A. sichuanensis* (holotype, *Wu 0010-18*).

numerous minute oily drops, colorless, 65–200 × 8.2–15.5 µm, thin- to slightly thick-walled, SA–. Acanthophyses numerous, clavate to broadly clavate, fusiform, stalked, colorless, apical parts with numerous protuberances, 50–100 × 5–30 µm, up to 1.2 µm thick walls, aculei 1–7 × 1–2 µm. Dendrohyphidia numerous, 37–90 × 3–4.8 µm. Hyphidia numerous, 35–80 × 2.4–4.2 µm. Basidia clavate, middle parts usually with several protuberances, 65–130 × 20–32 µm, up to 1.2 µm thick walls, 4-sterigmate. Basidiospores broadly ellipsoid to subglobose, adaxially flattened, aculeate, thin- to thick-walled, up to 3 µm thick walls, with a distinct apiculus, homogenous or with several oil-drops, amyloid, CB–, mostly 22.5–27.5 × 19–24 µm. (22.5–)23.5–27.2(–29) × (18.2–)19.2–22.8(–24) µm, L = 25.4±1.3 µm, W = 20.7±1.6 µm, Q = 1.23 (n = 30) (holotype, *Wu 1308-54*); (22.2–)23–26.5(–28) × (18.2–)20–22.5(–25.5) µm, L = 24.8±1.3 µm, W = 21.2±1.5 µm, Q = 1.17 (n = 30) (*Wu 1106-14*).

**Ecology and distribution.** On *Pinus* branches at high elevations, China and Taiwan, Jun to Aug.

**Additional specimens examined.** TAIWAN. Taichung, Siaosyueshan, Tienchih, 24°17'N, 121°01'E, 2580 m, on branch of *Pinus armandii*, 8 Jun 2011, *S.H. Wu*, *Wu 1106-14* (TNM F25532); *ibid. Wu 1106-16* (TNM F25534).

***Aleurodiscus senticosus* Sheng H. Wu, sp. nov.**
MycoBank MB823180
Figs 2C, 5

**Typification.** TAIWAN. New Taipei City, Wulai, 24°51'N, 121°33'E, 448 m, on branch of angiosperm, 10 Sep 2012, *S.H. Wu*, *Wu 1209-7* (holotype TNM F26702). GenBank: ITS = MH596849, 28S = MF043531, *TEF1* = LC271169.

**Etymology.** *senticosus* (L.) = full of thorns, referring to the surface of basidia and cystidia.

**Diagnosis.** Macroscopically featured in having a more or less cracked hymenophore, resulting from the fusion of numerous basidiome patches. Microscopically its basidia are diagnostic in having large lateral echinulate bladder-like swollen structure. Morphologically it resembles *Xylobolus* spp., although the latter cause a white-pocket rot in wood and have smooth basidiospores.

**Description.** Basidiomes resupinate, beginning as small orbicular patches, gradually extending and fusing together then becoming effused, adnate, membranaceous, 250–600 µm thick in section. Hymenial surface Buff or Light Buff, slightly tuberculate, with a more or less cracked hymenophore; margin paler, usually determinate, occasionally thinning and byssoid.

Hyphal system monomitic; hyphae simple-septate, colorless. Subiculum with dense texture, 200–350 µm thick; hyphae next to substrate more or less horizontal, slightly interwoven, colorless, moderately ramified, at the junction of basidiocarp patches more or less vertical, 2–4(–5) µm diam, walls up to 1.5 µm thick. Hymenial layer thickening, with dense texture, 150–250 µm thick, not clearly differentiated from the subiculum; hyphae mainly vertical, colorless, 2–4 µm diam, thin- to slightly thick-walled. Gloeocystidia numerous, immersed or slightly projecting, cylindrical or tubular, with stalked bases, apically sometimes forked, sometimes with one or more constrictions near apices or slightly moniliform, colorless, 45–135 × 5–12 µm, with walls up to 1.5 µm thick, SA–. Acanthophyses numerous, subclavate or clavate, basal parts thin-walled, thick-walled toward apices, colorless, median to apical parts echinulate, 25–65 × 4–13 µm (spines excluded). Hyphidia numerous, 35–65 × 2–4 µm. Basidia clavate, 60–82 × 10–15 µm, with walls up to 2 µm thick, 4-sterigmate, usually with large lateral echinulate bladder-like swollen structure. Basidiospores broadly ellipsoid to subglobose, adaxially flattened, aculeate, with 1–3 µm thick walls, homogeneous or sometimes with several oily drops, amyloid, CB–, mostly 13.5–16.5 × 11–13 µm. (13–)13.5–15.8(–17) × (10–)11.2–12.5(–13) µm, L = 14.8±1.00 µm, W = 11.8±0.6 µm, Q = 1.25 (n = 30) (holotype, *Wu 1209-7*); (13–)14–16(–17.2) × (10–)11.2–13(–15) µm, L = 15.1±1.0 µm, W = 11.9±1.0 µm, Q = 1.26 (n = 30) (*GC 1604-46*).

**Ecology and distribution.** On angiosperm branches, Taiwan, Apr to Sep.

**Additional specimens examined.** Taiwan, New Taipei City, Wulai, 24°51'N, 121°33'E, 448 m, on angiosperm branch, 10 Sep 2012, *S.H. Wu*, *Wu 1209-9* (TNM F26704); Nantou, Lienhuachih. 23°56'N, 120°53'E, 700 m, on angiosperm branch, 08 Oct 1996, *S.H. Wu*, *Wu 9610-1* (TNM F5344); on angiosperm branch, 09 Apr 2016, *G.C. Chen*, *GC 1604-46* (TNM F30771).

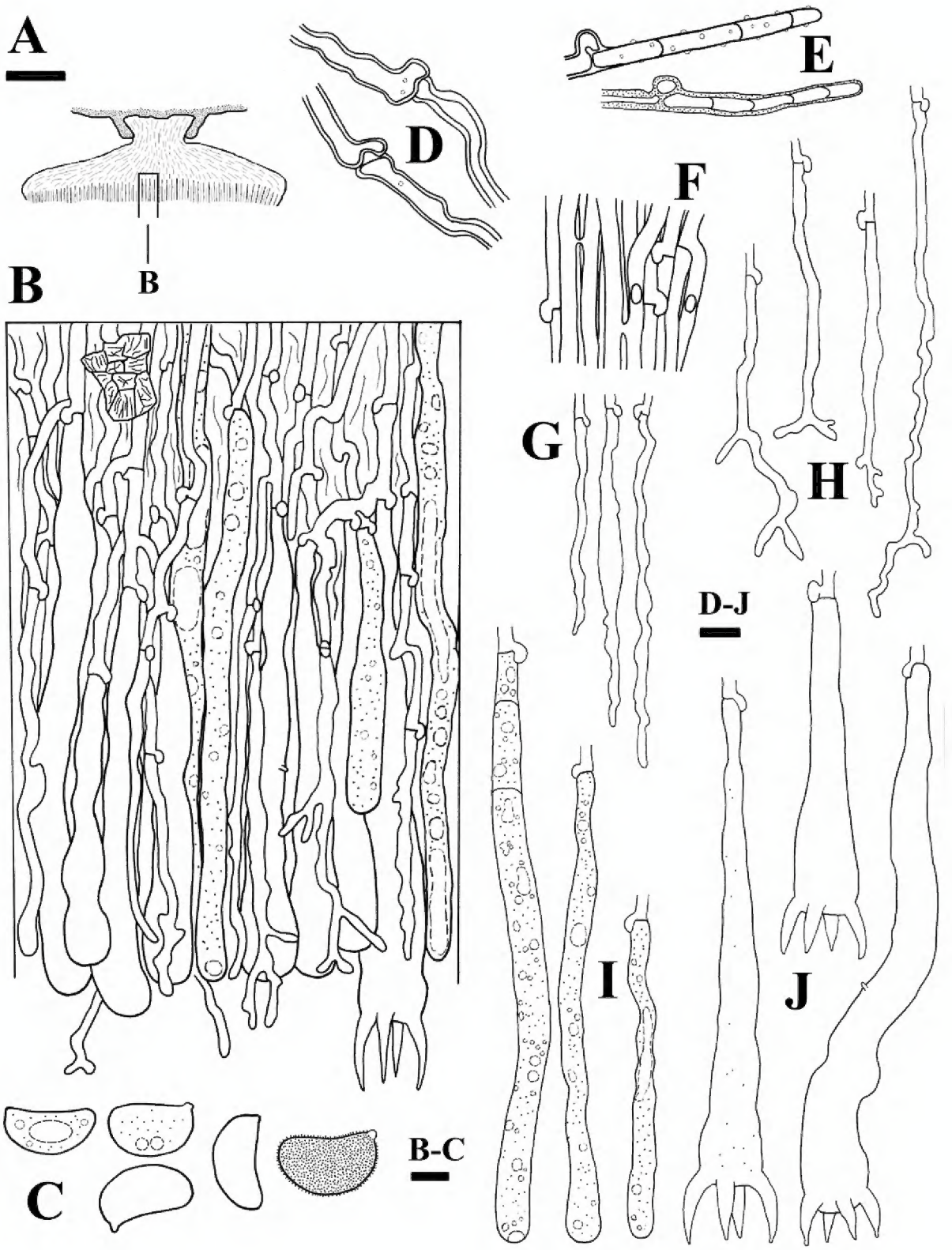


**Figure 3.** Microscopic structures of *Aleurodiscus alpinus* (holotype, *Wu 1407-55*) **A** profile of basidiocarp section **B** subhymenial and hymenial section **C** basidiospores (far right: in IKI) **D** subicular hyphae near substrate **E** pileus hyphae **F** subhymenial hyphae **G** hyphidia **H** branched hyphidia **I** gloeocystidia **J** basidia. Bars: 300 μm (**A**); 10 μm (**B–J**).

***Aleurodiscus sichuanensis*** **Sheng H. Wu, sp. nov.**
MycoBank MB823181
Figs 2D, 6

**Typification.** CHINA. SICHUAN PROVINCE: Wolungshan, Tengsheng, 2700 m, under bark of angiosperm, 11 Oct 2000, *S.H. Wu & S.C. Wu*, *Wu 0010-18* (holotype TNM F12097). GenBank: ITS = MH596852, 28S = MF043534, *TEF1* = LC269194.

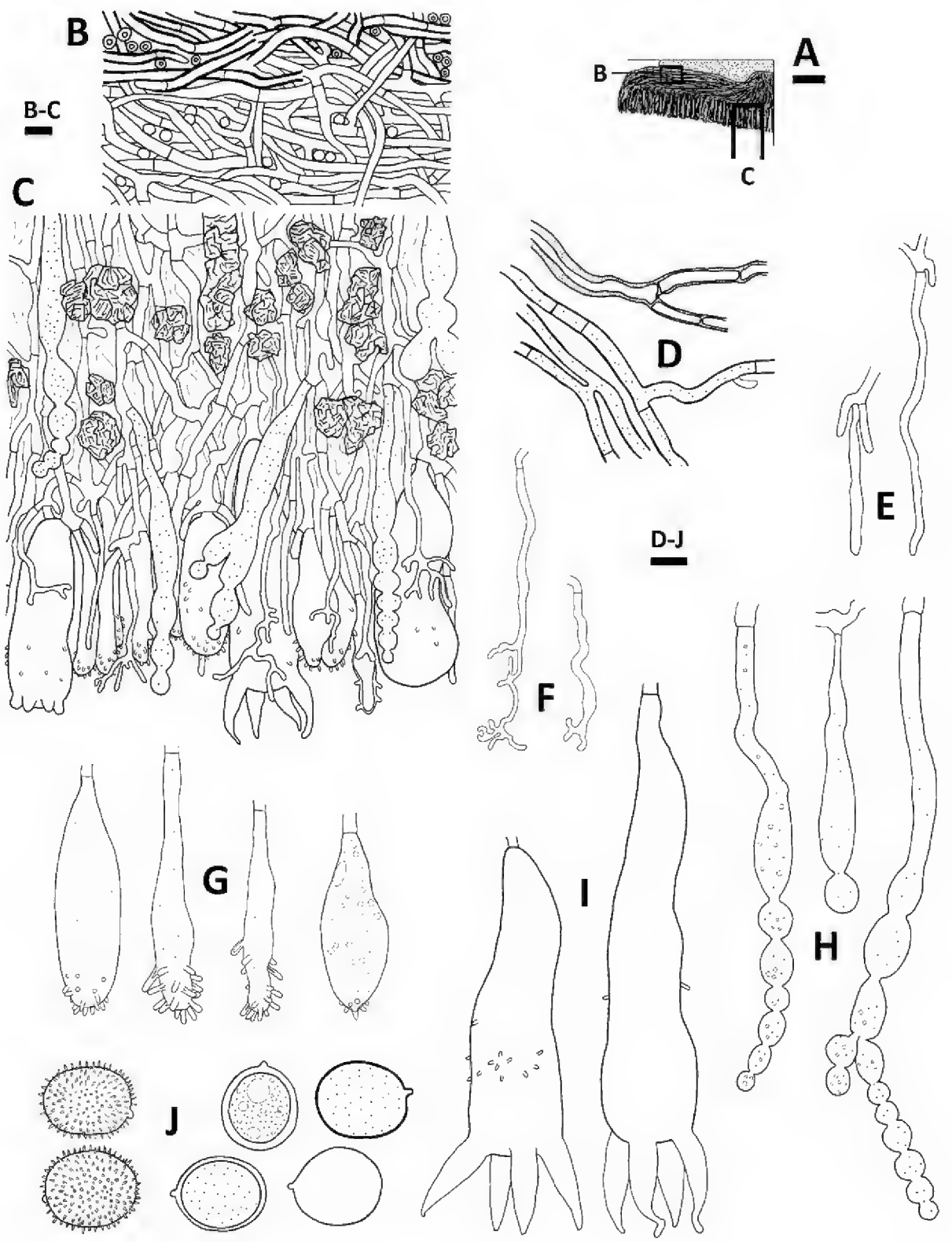


**Figure 4.** Microscopic structures of *Aleurodiscus pinicola* (holotype, *Wu 1308-54*) **A** profile of basidiocarp section **B** subicular hyphae of basidiocarp section **C** subhymenial and hymenial section **D** generative hyphae **E** hyphidia **F** dendrohyphidia **G** acanthophyses **H** gloeocystidia **I** basidia **J** basidiospores (left: in IKI, right: in KOH). Scale bars: 200 μm (**A**); 10 μm (**B–J**).

**Etymology.** *sichuanensis* (L.), referring to Sichuan Province, the type locality.

**Diagnosis.** *Aleurodiscus sichuanensis* resembles *A. oakesii* in having acanthophyses, simple-septate generative hyphae, and gloeocystidia occasionally with protuberances. However, clamped hyphae are rarely present in *A. oakesii*. Protuberances of acanthophyses of *A. oakesii* are antler-like, while aculei of acanthophyses in *A. sichuanensis* are fairly small. Basidiospores of *A. sichuanensis* are D-shaped or broadly ellipsoid, while those of *A. oakesii* are ovoid-ellipsoid and slightly smaller (18–27 ×12–14(–17) μm).

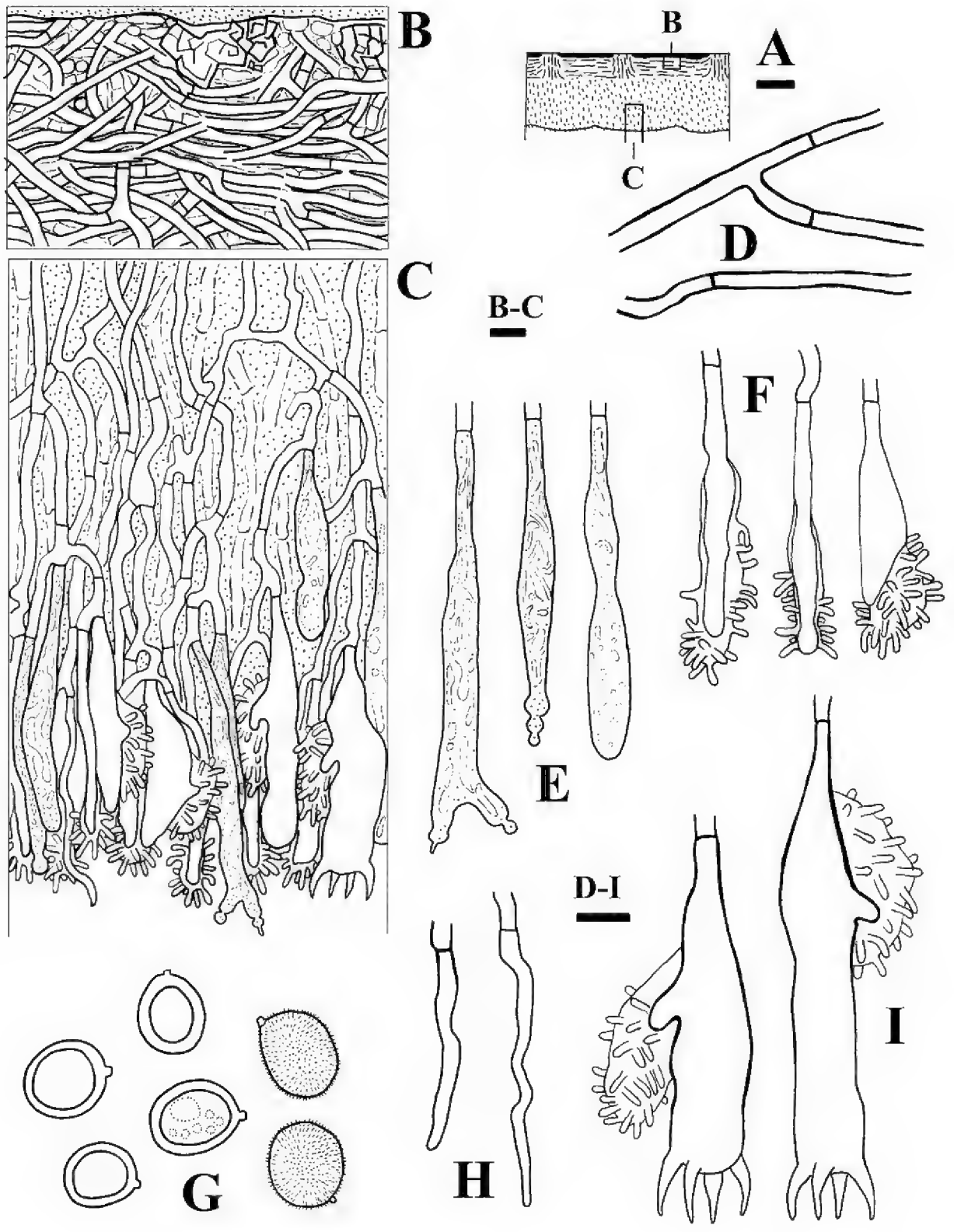


**Figure 5.** Microscopic structures of *Aleurodiscus senticosus* (holotype, *Wu 1209-7*) **A** profile of basidiocarp section **B** basal of basidiocarp section **C** section of hymenium **D** subicular hyphae **E** gloeocystidia **F** acanthophyses **G** basidiospores (left: in KOH, right: in IKI) **H** hyphidia **I** basidia. Scale bars: 200 μm (**A**); 10 μm (**B–I**).

*Aleurodiscus sichuanensis*, however, is most closely related to *A. alpinus* and differs from it by having acanthophyses and simple-septate hyphae.

Basidiomes resupinate, effused, adnate, membranaceous-subceraceous, 150–350 μm thick in section. Hymenial surface smooth, Buff or Buff Yellow, occasionally cracked; margin concolorous, determinate.

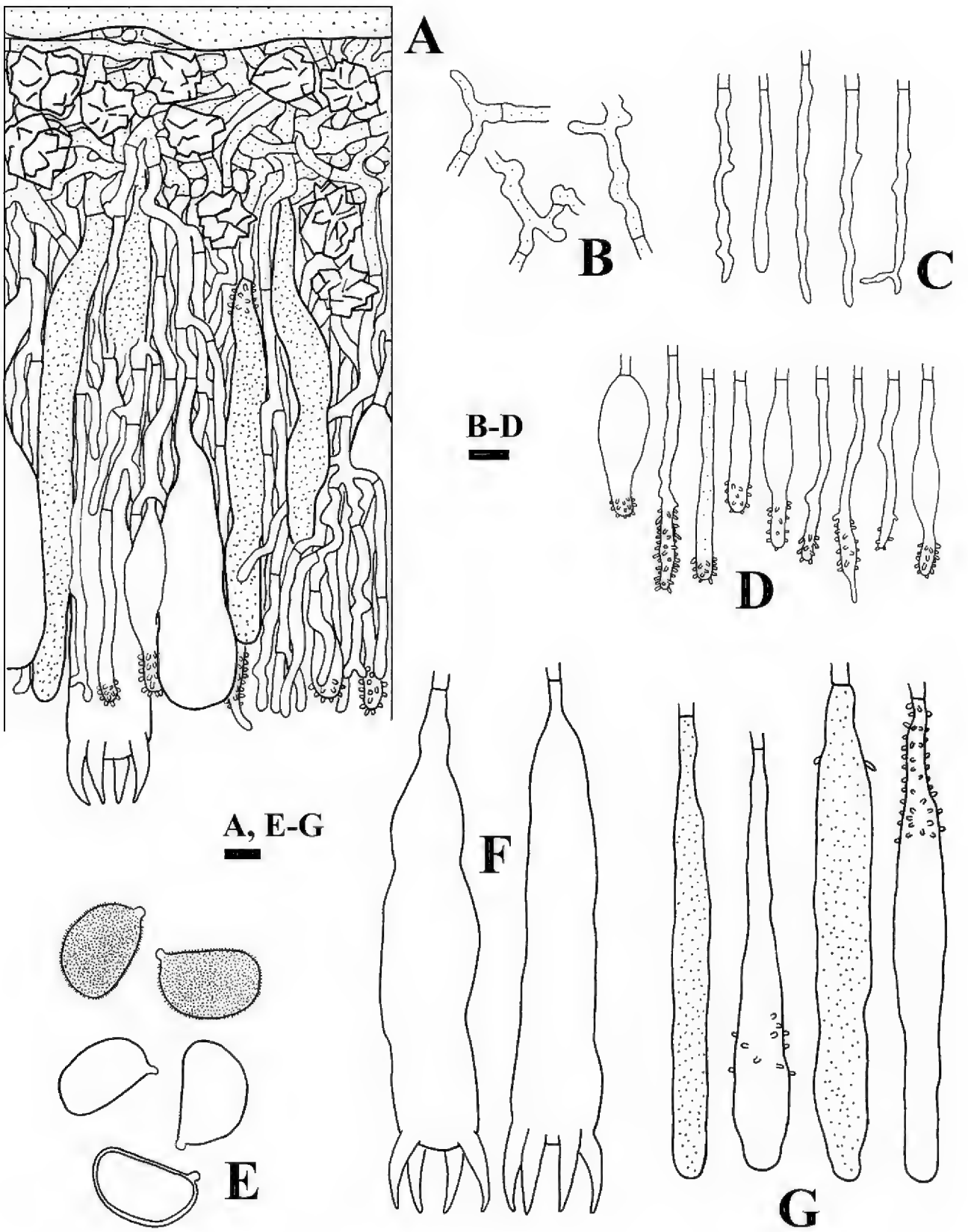


**Figure 6.** Microscopic structures of *Aleurodiscus sichuanensis* (holotype, *Wu 0010-18*) **A** basidiocarp section **B** subicular hyphae **C** hyphidia **D** acanthophyses **E** basidiospores (upper: in IKI, lower: in KOH) **F** basidia **G** gloeocysitidia. Scale bars: 10 μm.

Hyphal system monomitic; hyphae simple-septate. Subiculum uniform, with dense texture, thin or up to 150 μm thick; hyphae interwoven, colorless, richly ramified, tortuous, usually full of small oily drops, 2.5–5.5 μm diam, thin-walled. Hymenial layer with dense texture, 100–200 μm thick; hyphae vertical, colorless, ± straight, 2.5–4.5 μm diam, thin-walled. Crystal masses scattered in subiculum, yellowish. Gloeocystidia

numerous, immersed or projecting, yellowish or pale brownish yellow, cylindrical, narrowly clavate or tubular, with oily contents or homogeneous, SA+, basal or median portion occasionally with small aculei, 70–135 × 7–14 µm, with 0.5–1 µm thick walls. Acanthophyses numerous, irregularly cylindrical or narrowly clavate, sometimes subfusiform, colorless, apical parts with numerous aculei, 30–70 × 3–8(–12) µm (aculei excluded), thin-walled. Hyphidia numerous, occasionally branched, 35–85 × 2.5–4.5 µm. Basidia clavate, 4-sterigmate, 100–130 × 20–25 µm, with 0.8–1.2 µm thick walls. Basidiospores D-shaped or broadly ellipsoid, adaxially flattened, finely aculeate, thin-walled or 1–2 µm thick, sometimes with oily contents, amyloid, CB–, mostly 25.5–28.5 × 15–18 µm. (25–)26–28.2(–29) × (14.5–)15.2–17(–19) µm, L = 27.1±1.0 µm, W = 15.9±1.1 µm, Q = 1.71 (n = 30) (holotype, *Wu 0010-18*).

**Ecology and distribution.** On dead branches of *Quercus* and other angiosperms at high elevations, China, Jul to Oct.

**Additional specimens examined.** CHINA. SICHUAN PROVINCE: Wolungshan, Tengsheng, 2700 m, on angiosperm branch, 11 Oct 2000, *S.H. Wu & S.C. Wu, Wu 0010-42* (TNM F12118). YUNNAN PROVINCE: Shangrila County, Pudacuo National Park, 3600 m, on dead branch of *Quercus apuifolloides*, 28 Jul 2017, *S.H. He, He 4923*, *He 4926*, *He 4930*, *He 4935* (BJFC).

## Discussion

A number of phylogenetic studies of *Aleurodiscus* s.l. have been conducted in the past twenty years (Wu et al. 2001; Larsson and Larsson 2003; Miller et al. 2006; Larsson 2007; Dai and He 2016; Dai et al. 2017). Miller et al. (2006) and Larsson (2007) tried to establish a family level classification for *Aleurodiscus* s.l., as well as related taxa of the Russulales. However, a fully resolved and robust phylogeny of *Aleurodiscus* s.l. and related taxa was not achievable with ribosomal genes alone. Dai and He (2016) and our study have addressed this by including *TEF1* for phylogenetic analyses. From our phylogenetic analyses of three DNA genetic markers (Fig. 1) we can conclude the following about evolutionary relationships in the Stereaceae: (i) *Aleurodiscus* s.l. is highly polyphyletic; (ii) *Acanthophysellum* is polyphyletic; (iii) *Gloeocystidiellum* is polyphyletic; (iv) *Megalocystidium* is polyphyletic; and (v) *Conferticium* is paraphyletic.

*Aleurodiscus alpinus* is reminiscent of *Aleurodiscus* s.s. (*A. amorphus* (Pers.) J. Schröt. and *A. grantii* Lloyd) due to the discoid basidiocarp and echinulate basidiospores, as well as the absence of acanthophyses. However, the gloeocystidia of *Aleurodiscus* s.s. are paraphysis-like, narrow and moniliform, while those of *A. alpinus* are much wider and not moniliform. In addition, *A. alpinus* has unbranched or branched hyphidia, which are lacking in *Aleurodiscus* s.s. *Aleurodiscus alpinus* formed a clade with *A. sichuanensis* (Fig. 1), however, the latter has simple-septate hyphae and acanthophyses. *Aleurodiscus alpinus* and *A. cupulatus* share most morphological features, except the latter has much wider basidiospores. *Aleurodiscus alpinus* grows on *Rhododendron* sp. in Yunnan of

China, while *A. cupulatus* occurs on *Pseudotsuga menziesii* in Idaho of USA. No DNA sequence of the latter has been obtained to examine their relationship.

*Aleurodiscus pinicola* presents protuberances in the basidia and this is reminiscent of *Acanthobasidium*. However, this feature is not limited to *Acanthobasidium* spp. For example, basidia of *Aleurodiscus mirabilis* (Berk. & M.A. Curtis) Höhn. and *A. wakefieldiae* Boidin & Beller occasionally possess protuberances, but they and *A. pinicola* do not belong to *Acanthobasidium* (Fig. 1).

*Aleurodiscus senticosus* is macroscopically distinct in having more or less cracked hymenophore from the fusion of smaller basidiocarp patches; microscopically, its basidia bear a large, spiny, bladder-like structure that is unique among *Aleurodiscus* s.l. The present phylogenetic analyses (Fig. 1) indicated that *A. senticosus* formed a clade with *Xylobolus* and *Acanthofungus*, but without strong support. However, these two genera differ from *A. senticosus* by causing a white-pocket rot in wood and by bearing smooth basidiospores.

*Aleurodiscus sichuanensis* cannot be accommodated in any segregate genus of *Aleurodiscus* s.l., according to the combined features of effused basidiocarp, simple-septate hyphae, acanthophyses, gloeocystidia with aculei, and echinulate basidiospores.

In conclusion, the status of each segregate genus of *Aleurodiscus* s.l. should be further examined by multi-gene analysis of more species to evaluate which ones can be recognized and which cannot. Although the four new species we introduce cannot be accommodated in any segregate genus of *Aleurodiscus* s.l. according to the present combined morphological and phylogenetic studies, they are still placed under the broad sense of *Aleurodiscus* at the present time.

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